PB86-155777

Plant Exposure Laboratory and Chambers. Volume 1

(U.S.) Corvallis Environmental Research Lab., OR

Jan 86

U.S. Department of Commerce
National Technical Information Service
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PB86-155777

EPA/600/3-86/007a
January 1986

PLANT EXPOSURE LABORATORY AND CHAMBERS

Volume I.

by

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TECHNICAL REPORT DATA (Please read Instructions on the reverse before completing)		
1. REPORT NO. EPA/600/3-86/007a	2.	3. RECIPIENT'S ACCESSION NO. PB86 155777/AS
4. TITLE AND SUBTITLE PLANT EXPOSURE LABORATORY AND CHAMBERS: Volume I.		5. REPORT DATE January 1986 6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S) C.McFarlane, T. Pfleeger, M. McDowell, J. Wilson and R. Shimabuku		8. PERFORMING ORGANIZATION REPORT NO.
9. PERFORMING ORGANIZATION NAME AND ADDRESS ERL-Corvallis		10. PROGRAM ELEMENT NO. 11. CONTRACT/GRANT NO.
12. SPONSORING AGENCY NAME AND ADDRESS Environmental Research Laboratory Office of Research and Development U.S.Environmental Protection Agency Corvallis, OR 97333		13. TYPE OF REPORT AND PERIOD COVERED In-house Report 14. SPONSORING AGENCY CODE EPA 600/02
15. SUPPLEMENTARY NOTES 1985. Volume II of this report contains Appendices 1 thru 6		

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17. KEY WORDS AND DOCUMENT ANALYSIS		
a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
18. DISTRIBUTION STATEMENT Release to Public	19. SECURITY CLASS (This Report) Unclassified	21. NO. OF PAGES 35
	20. SECURITY CLASS (This page) Unclassified	22. PRICE

EPA Form 2220-1 (9-73)

DISCLAIMER

The information and research described in this document has been funded wholly or in part by the United States Environmental Protection Agency. It has been subjected to the Agency's peer and administrative review, and it has been approved for publication as an EPA document. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

ABSTRACT

This paper describes the laboratory and support system, the exposure chambers, the computer system, and the hydroponic nursery built to accomplish research on uptake, translocation, and metabolism of toxic organic chemicals by plants. Understanding these processes and the physiological driving mechanisms is essential to predict food chain contamination and environmental damage from various agricultural and industrial pollutants. Since the chemicals of interest are toxic and studies generally include ^{14}C as a label for monitoring chemical kinetics, containment was an important criterion. Containment and safety considerations are included.

CONTENTS

	Page
Abstract	iii
Figures	vi
Tables	vi
Acknowledgments	vii
Introduction	1
Plant Exposure Laboratory	1
Exposure Chambers	2
Shoot Compartment	4
Root Compartment	11
Hydroponic System	11
Soil System	13
Computer System (Control and Data Collection)	14
Hardware	14
Software	16
Hydroponic Nursery	18
Performance	18
Continuously Stirred Tank Reactor (CSTR)	18
Mixing	18
Turbulence	20
Photosynthesis	20
Environmental and Plant Conditions	21
Literature Cited	26

Appendices

1. Control Program: User's Guide
2. Control Program: Listing and Description
3. Plant Exposure Chamber Construction Drawings and Parts List
4. Electronic Diagrams for Components of Plant Exposure Laboratory
5. Thermistor Construction and Calibration
6. Hydroponic Nursery and Nutrient Formulations

FIGURES

Number		Page
1	Plant exposure laboratory floor plan	viii
2	Plant exposure laboratory elevation	3
3	Support connections for plant exposure chamber	3
4	Continuously stirred tank reactor (CSTR)	4
5	Condenser and transpired water collector	5
6	Air movement past the condenser	6
7	Hydroponic plant pots	7
8	Air sampling schematic	9
9	Transpiration rate measurement	10
10	Solution sampling port	11
11	Instrumentation pot	12
12	Soil pot placement detail	13
13	Hydroponic plant nursery	19
14	CSTR mixing rate	20
15	Air turbulence in the CSTR	21
16	Automatic CO_2 control	23
17	Computer display of measured parameters	24

TABLES

1	Measurement instrument locations and recorded units	14
2	Computer specifications and electronic components	15
3	Program routines and functions	16
4	Automatically controlled function on each plant exposure chamber	17
5	Statistics screen display	25

ACKNOWLEDGEMENTS

The concept of this laboratory and plant exposure system was born of need and made possible by the advent of cheap computers and advanced electronic devices. Support and encouragement for its completion has come primarily from Dr. Harold Kibby (Chief, Toxics and Hazardous Materials Branch) and Dr. Thomas Murphy (Laboratory Director).

John Wilson (Bendix Field Engineering Corporation) designed and built many of the electronic interfaces to the computer and all of the measurement and control circuits for the plant exposure chambers. He was responsible for selecting the commercial electronic components and for wiring the entire system. Without his dedication, this project could not have been accomplished.

Michael McDowell created the computer programs which manage the laboratory, collect the data and analyze and display the results. He was helped by others in the development of certain segments. Machine language routines were written by Allen Todd. Some segments were developed by Brian Martin and others by Robert Clark.

Design, construction, and testing of this laboratory and the plant exposure chambers, as well as the writing of this description has been a group effort. This includes valuable contributions by Donald Craig, Amy Rowe, Katy Carmichael, Renee Harber, Kim Hayden, Raymond Shimabuku, and Mark Champlin. Various aspects of construction were done by employees of the Bendix Field Engineering Corporation. Especially important was the work by Walter Guetter and John Friedrichsen, and consultation, suggestions and support by Robert Hazelton.

Illustrations were done by Ronald Selig, Brennan Raebel, and Jeffery Nyquist.

The Duke University Phytotron (Dr. Boyd Strain, Director, and Newton McQuay, Engineer) made available two fluorescent light units which are used for lighting one of the controlled environment areas in the laboratory.

Layout and editing was done by Karen Randolph; Carol Roberts typed the manuscript and appendices.

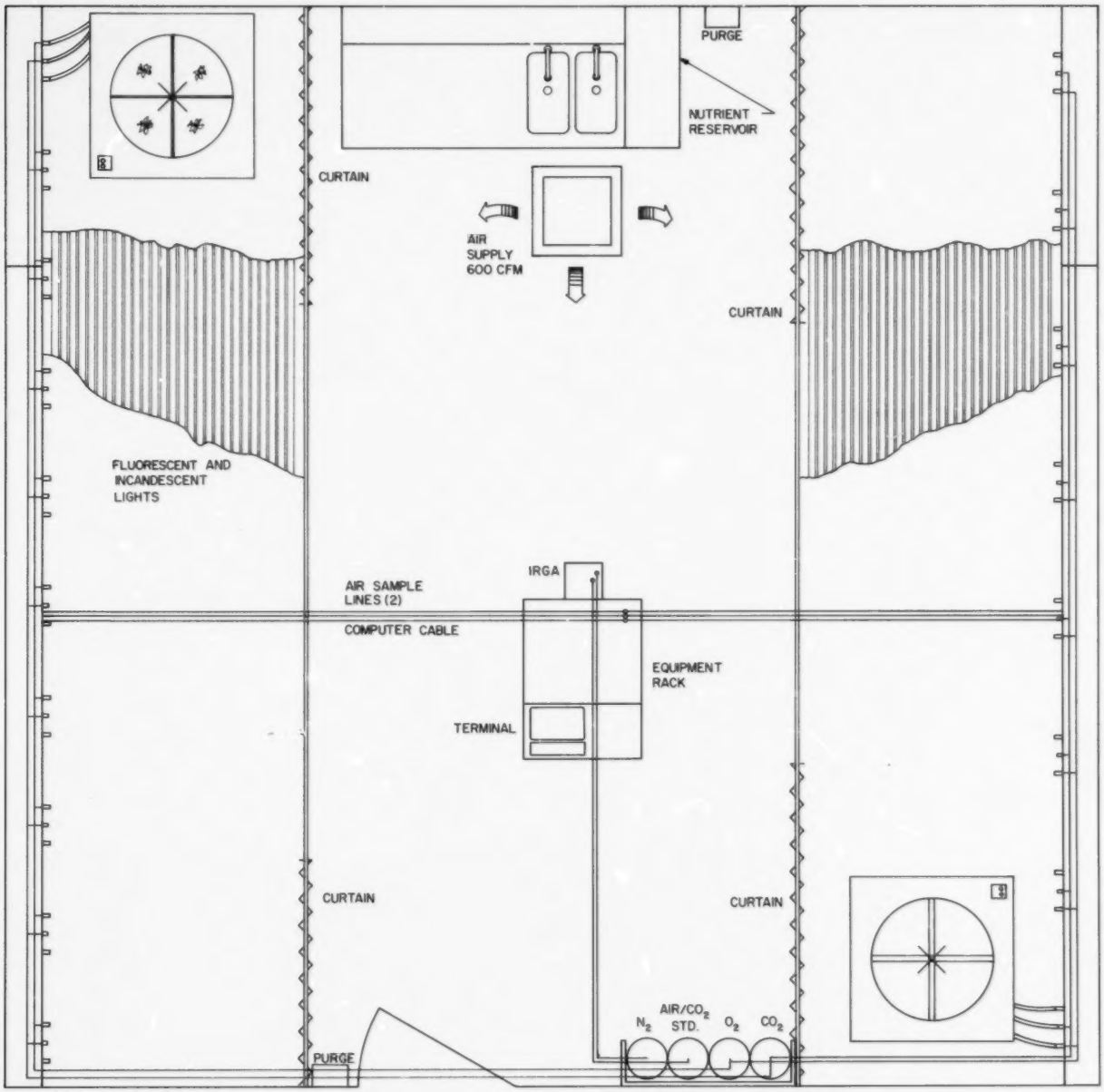


Figure 1. Plant exposure laboratory floor plan.

INTRODUCTION

Our research is to learn the factors which control plant uptake, translocation and metabolism of anthropogenic organic chemicals. Understanding these processes is essential to predict food contamination and environmental damage from various agricultural and industrial pollutants. Contamination of plants is only one component, but since plants are the fulcrum upon which all nourishment systems depend, understanding the ways they become contaminated is critical to prudent production, transportation and use of organic chemicals.

These efforts to identify the controlling mechanisms of these phenomena require an understanding of the physiological parameters of the plants during uptake and translocation of the extraneous chemicals. Since the chemicals of interest are toxic and studies generally include ^{14}C as a label for monitoring chemical kinetics, containment is an important criterion. This paper describes the laboratory and support system, the exposure chambers, the computer system, and the plant hydroponic nursery built to accomplish this research.

PLANT EXPOSURE LABORATORY

The plant exposure laboratory provides a controlled environment for plant physiology research in which toxic pollutants can be contained and studied. It is built to accommodate movable plant exposure chambers within a temperature and light controlled area.

The laboratory is divided into three sections (Fig. 1). The east and west portions are separated from the center by white opaque curtains which hang to within 4 cm of the floor and create two growth chambers, approximately 12 m^2 each. The curtains are suspended from a track and can be pulled open from the middle or either end, allowing easy access to any portion of the controlled area.

Heat exchange units in each area provide cooling by moving air in a circular pattern through adjustable registers to create uniform circulation throughout the controlled area (Fig. 2). Temperatures are adjustable between 15C and 40C (light) and 15C to 25C (dark). Two light units (4 total) are hung end to end against each wall. Each contains 24 8-foot, 1500 ma deluxe cool white fluorescent lamps and 10 67-watt clear incandescent lamps. The light fixtures are kept at a constant temperature by circulated air which passes a watercooled radiator. A plexiglass barrier separates the lamps from the controlled area. The average photosynthetically active radiation (PAR) at plant height is 350 µmol s-1 m-2.

The center of the room contains work benches, a sink, the microcomputer and other electronic and air sampling equipment. This space is used for sample preparation, analysis and other activities associated with the lab.

Temperature controlled air from the building heating/cooling system (17 m3 min-1) enters the center work area, flows under the curtains into the control areas and exhausts to the outside. This is a safety system which assures clean air for workers and prevents contaminated air from entering other parts of the building in the event of an accident.

Plant exposure chambers connect to the support system at any of 18 positions. Plug-in supports are positioned along the wall (Fig. 3) and include: 115 VAC, 100-wire computer connection, chamber gas sampling and return, nutrient solution, CO_2, O_2, a vacuum line, and supply and return of a controlled temperature coolant (generally 2C).

The air contained within each plant exposure chamber will at times contain toxic gases. Prior to opening such a chamber the gas must be removed. This is accomplished by attaching a vacuum (260 l m-1) to a vent on the chamber and opening another vent to allow replacement with ambient air. A vacuum outlet (Sew-Vac model I) is located on the north and south walls. From these outlets, a hose can reach each exposure chamber within the room. Air from the exposure chambers is passed through a filter and exhausted out the rear of the building.

EXPOSURE CHAMBERS

Two plant exposure chamber designs are available. One, designed for volatile gas uptake studies (Mc Farlane et al. 1981), was modified to work in the plant exposure laboratory. It has previously been described and won't be detailed herein. There are two variations of the other chamber, one for hydroponic and one for solid rooting materials (soils). Both use the same shoot compartment and differ only in the root support system. The air compartment contains the leaves and stems and provides a controlled environment in which the normal plant functions (i.e., photosynthesis, transpiration, and respiration) are monitored and controlled. The root compartment contains the roots in a manner to allow normal respiration, transpiration, and nutrient and anthropogenic chemical uptake. Detailed drawings and a parts list are included in Appendix 3. All chambers use the same electronic package and connections.

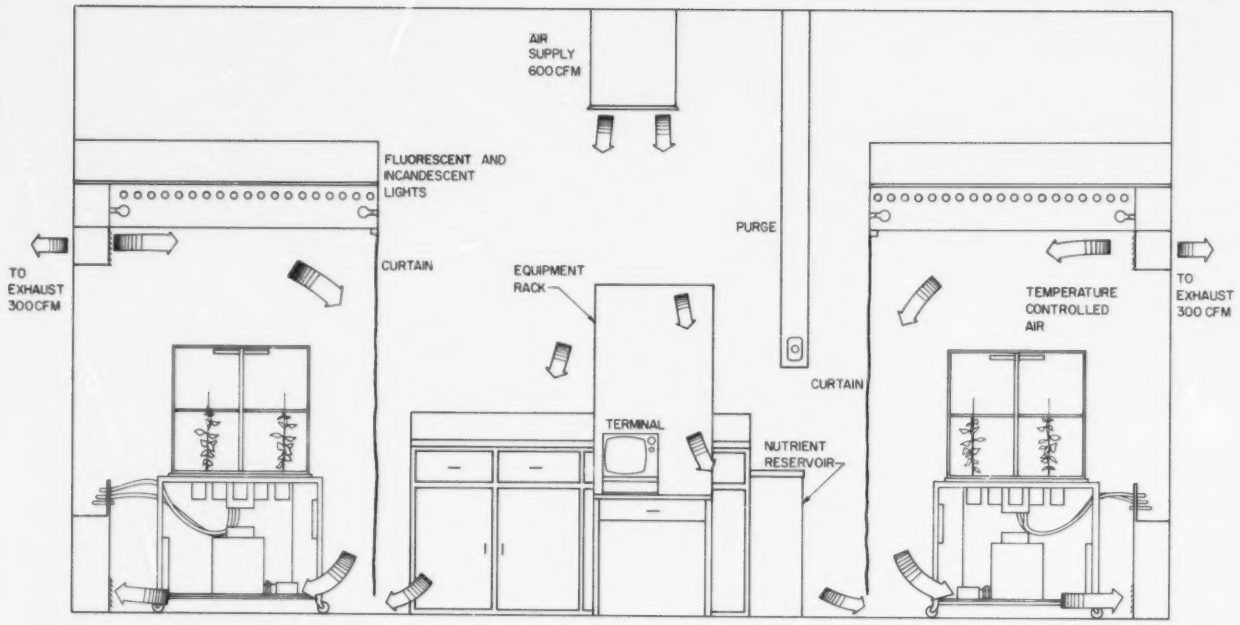


Figure 2. Plant exposure laboratory elevation showing air movement patterns, computer, controlled environments on each side and the chamber connection panel.

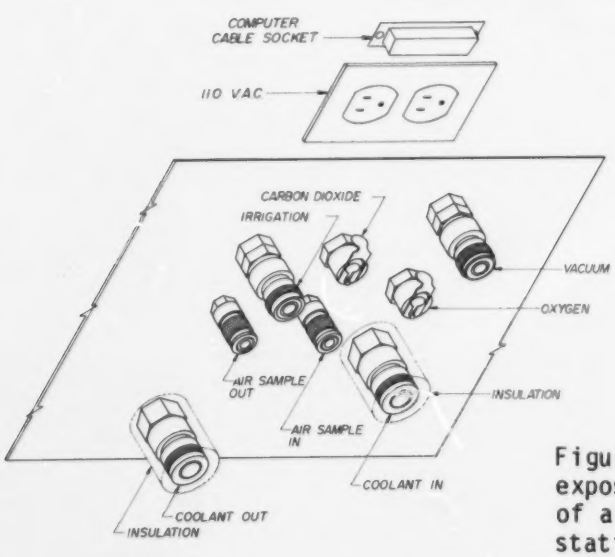


Figure 3. Support connections for plant exposure chamber. Shown is one segment of a two-part panel which consists of 10 stations on the east and 8 on the west wall.

The computer is initialized with identifying information which allows correct management. Differences such as chamber volume (used in photosynthesis calculations) or nutrient solution disperal (e.g., dripped onto a medium or added to a tank) are accommodated by the program.

SHOOT COMPARTMENT

The shoot compartment (Fig. 4) was constructed with an aluminum framework covered with a teflon skin. It is sealed to the table top by pressure clamps and a teflon-covered foam gasket. Air turbulence is created by a variable speed impeller mounted in the top of the compartment. This creates air movement at the leaf surfaces which is necessary for plant reactions and provides an ideal chamber for gas dynamic studies. The chamber meets the criterion of a continuously stirred tank reactor (CSTR) (Rogers et al. 1979). Air turbulence is a function of voltage to the impeller motor.

Air circulates by a variable speed fan from the chamber through a cold water condenser which condenses atmospheric humidity. Condensed water is drained away from the condenser and collected (Fig. 5). The air circulation rate can

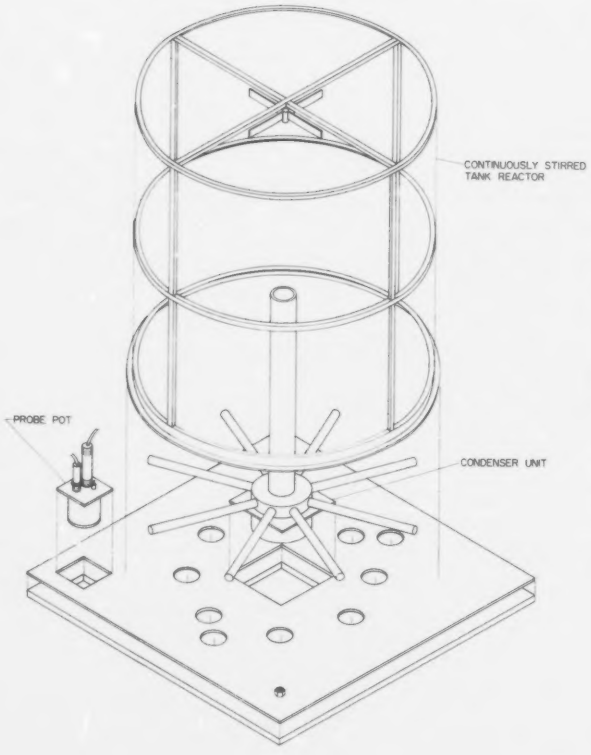


Figure 4. Continuously stirred tank reactor (CSTR). The tank is made of an aluminum frame covered with teflon and secured to the table with clamps. The plant pots are shown as holes on the table surface and the instrument pot is outside the CSTR. The condenser with air collection tentacles and return air tower are also shown in this exploded view. The impeller (for air mixing) is connected to the shaft of a 12 VDC explosion-proof motor which is mounted at the top of the tank.

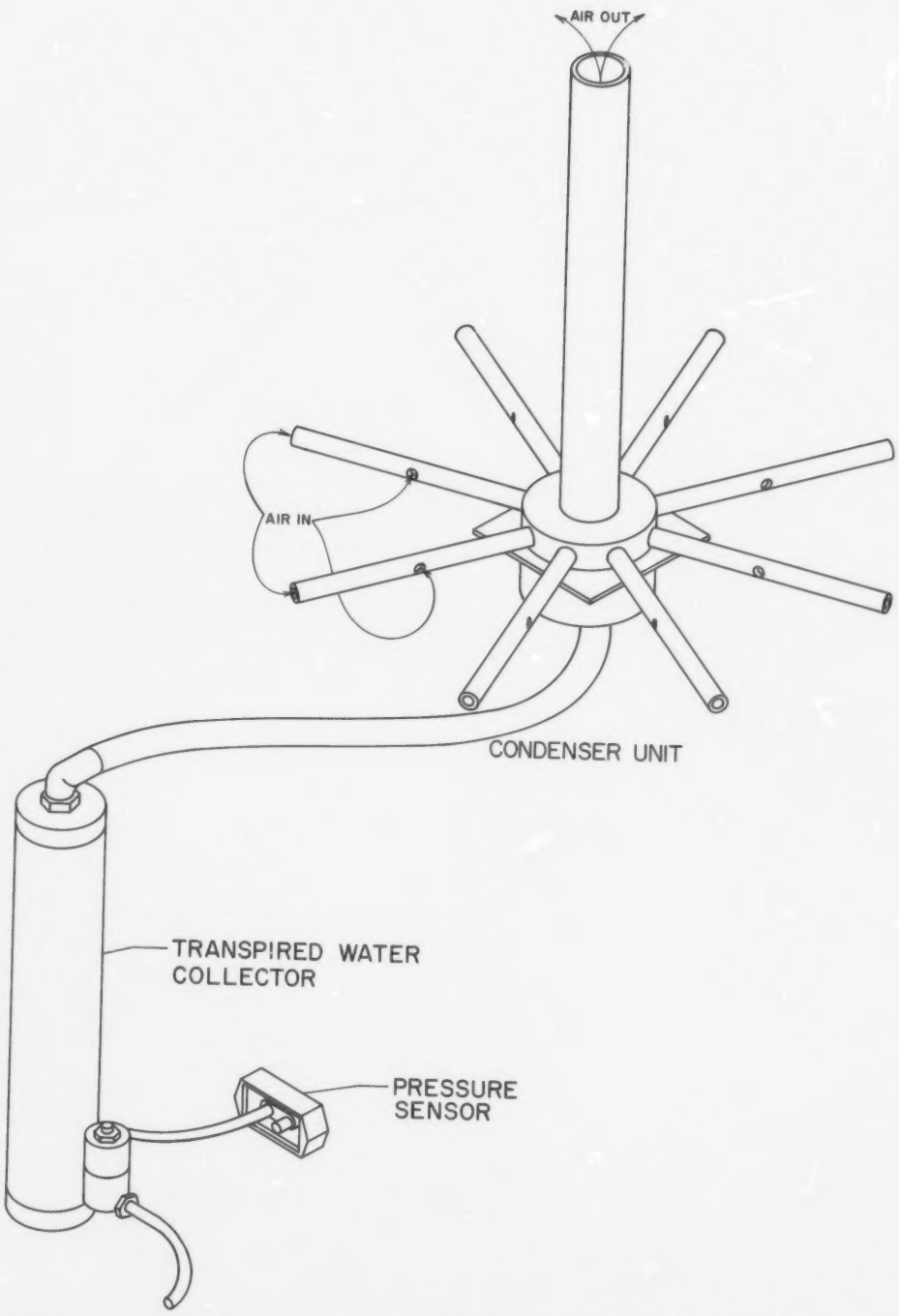


Figure 5. Condenser and transpired water collecter. A 12 VDC fan draws air from the chamber through 8 tentacles with open ends and median holes, past a chilled condenser and back to the chamber. Precipitated water drains to the collector where the transpiration rate is determined by periodically measuring the water height with a pressure sensor.

be controlled by changing the voltage to the fan (Fig. 6). Humidity in the chamber can be regulated within limits by changing the condenser temperature, the fan speed and the number or size of plants.

Changes in the condenser temperature and the air circulation fan speed have little effect on the chamber air temperature. The teflon cover allows temperature exchange and thus the temperature of the controlled environment room is primarily responsible for the chamber air temperature.

Separation between the shoot (top) compartment and the root (bottom) compartment (Fig. 7) consists of a plant support that is made of Drisco material which fits into a recess in the hydroponic pot. It is sealed in place with Teflon pressure-sensitive tape. The stem of the plant is held in place and sealed by a piece of closed-cell polyethylene foam. This allows the stem to expand without breaking the seal or damaging the plant as it grows.

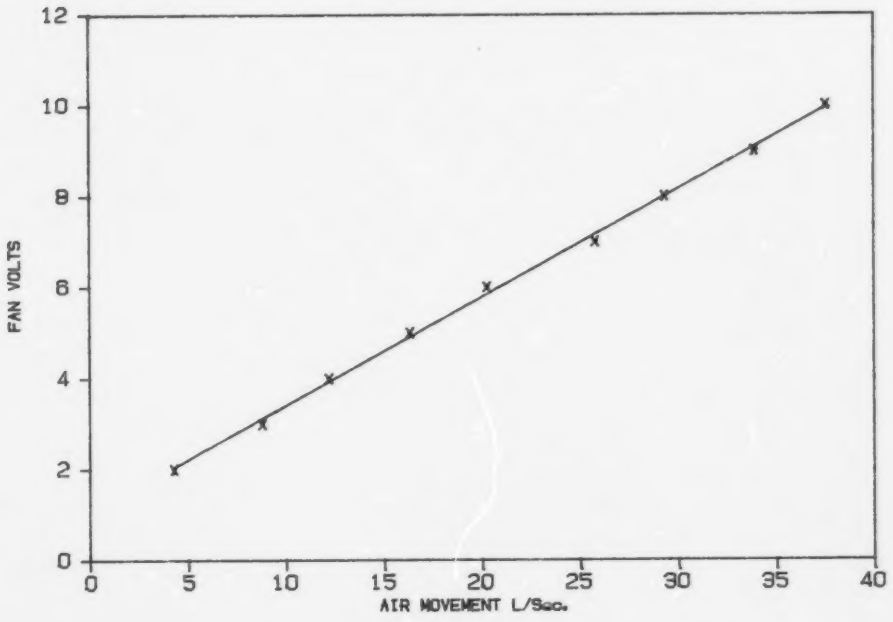


Figure 6. Air movement past the condenser is controlled by the voltage applied to the circulation fan. Normally the fan is adjusted to 6 V.

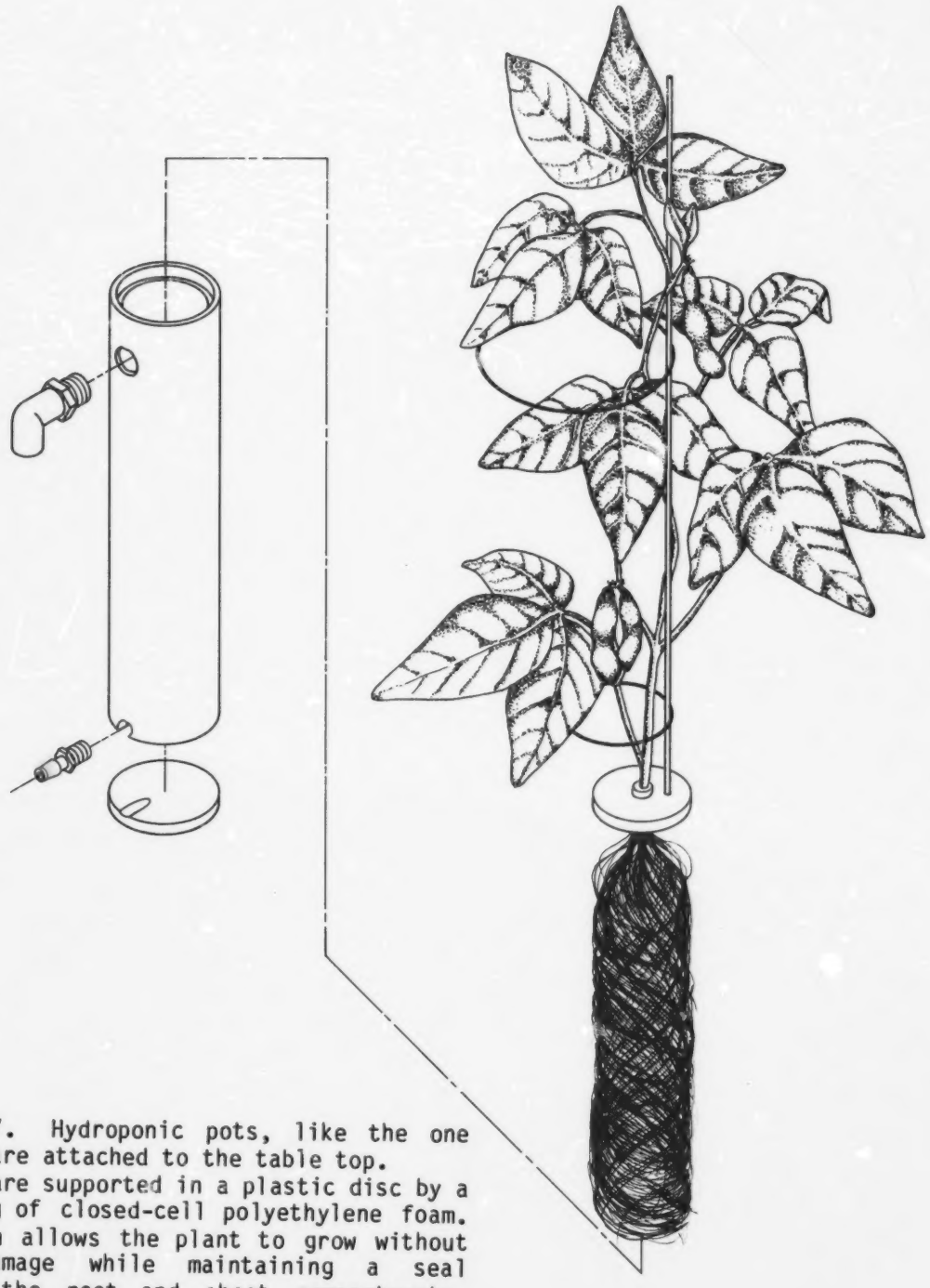


Figure 7. Hydroponic pots, like the one shown, are attached to the table top. Plants are supported in a plastic disc by a wrapping of closed-cell polyethylene foam. The foam allows the plant to grow without stem damage while maintaining a seal between the root and shoot compartments. The plant is supported by a stainless steel rod attached to the disc. The disc fits into a recess in the pot and is sealed to the table with teflon tape.

Air is sampled for CO_2 and H_2O by a closed loop gas sampler (Fig. 8). This is controlled by the microprocessor (Apple //e) which works in the following sequence: (1) Purge of the sample line, pump, and analytical equipment with outside air for 30 seconds. This is done by activating a pump and two valves, one of which brings outside air into the system via 1/4-inch polyethylene tube. The other vents the gas through a series of filters and then exhausts it outside; (2) Activate a valve pair for the chamber of interest, then pump air from that chamber at 0.800 l min-1 to fill all lines a nd analytical equipment. Equilibrium is achieved after about 20 seconds but the program waits 30 seconds and then (3) records measurements of CO_2 and H_2O concentrations from the infrared gas analyzer (IRGA); (4) Return to purge cycle; (5) Select next chamber and repeat the cycle. Periodically a standard gas (CO_2 in air) and a zero standard (N_2) are measured and the values are recorded.

Two important aspects were considered in designing the closed air system. One was to provide containment of gaseous pollutants or gaseous metabolic products. The second was to allow control and measurement of the plants' physiology, in particular photosynthesis, transpiration, and respiration.

Net CO_2 assimilation is controlled by injecting CO_2 into the chamber at a rate equivalent to the utilization rate at the desired concentration. This is accomplished by an electronic gas flow controller which is adjusted by the computer. The program makes the adjustment on the basis of the current CO_2 concentration, the concentration measured at the last sampling and the desired concentration. The logic and program are described in Appendix 2.

Figure 8 (page 9). Air sampling schematic. Gas is sampled from each chamber sequentially when the computer activates a pair of valves on manifolds (1) and (2) and controls the three-way valves (3 and 3A) to select from the east or west controlled area. The gas is pumped in a closed loop to and from the chamber through a 1/8-inch O.D. heated stainless steel tube by a stainless steel bellows pump at 0.800 l min-1. Gas samples are diverted by a manifold (4) through various measuring instruments. The flow to each is controlled by a rotometer (5) and collected into another manifold (6) for return to the plant exposure chamber. The bypass (7) returns air not needed for measurement. The unused ports on manifolds (4) and (6) are intended for future studies when an automatic sampling gas chromatograph and a flow-through internal proportional ^{14}C, ^{3}H analyzer will be used. CO_2 can be injected into the returning gas sample, manually by an electric toggle switch or by a computer-operated switch, either of which open valve (8). This system is used to bring the chamber up to the control CO_2 concentration when conditions have resulted in a value lower than desired. See the CO_2 control logic (Appendix 2, AIRSTUFF) for an explanation of the automatic use. Precise injection volumes depend on a calibrated capillary (0.005 inch I.D., 10.2 cm long) (9), regulated pressure (10), and precise timing which is calculated and controlled by the computer. The gauge is to check and allow adjustment of CO_2 pressure.

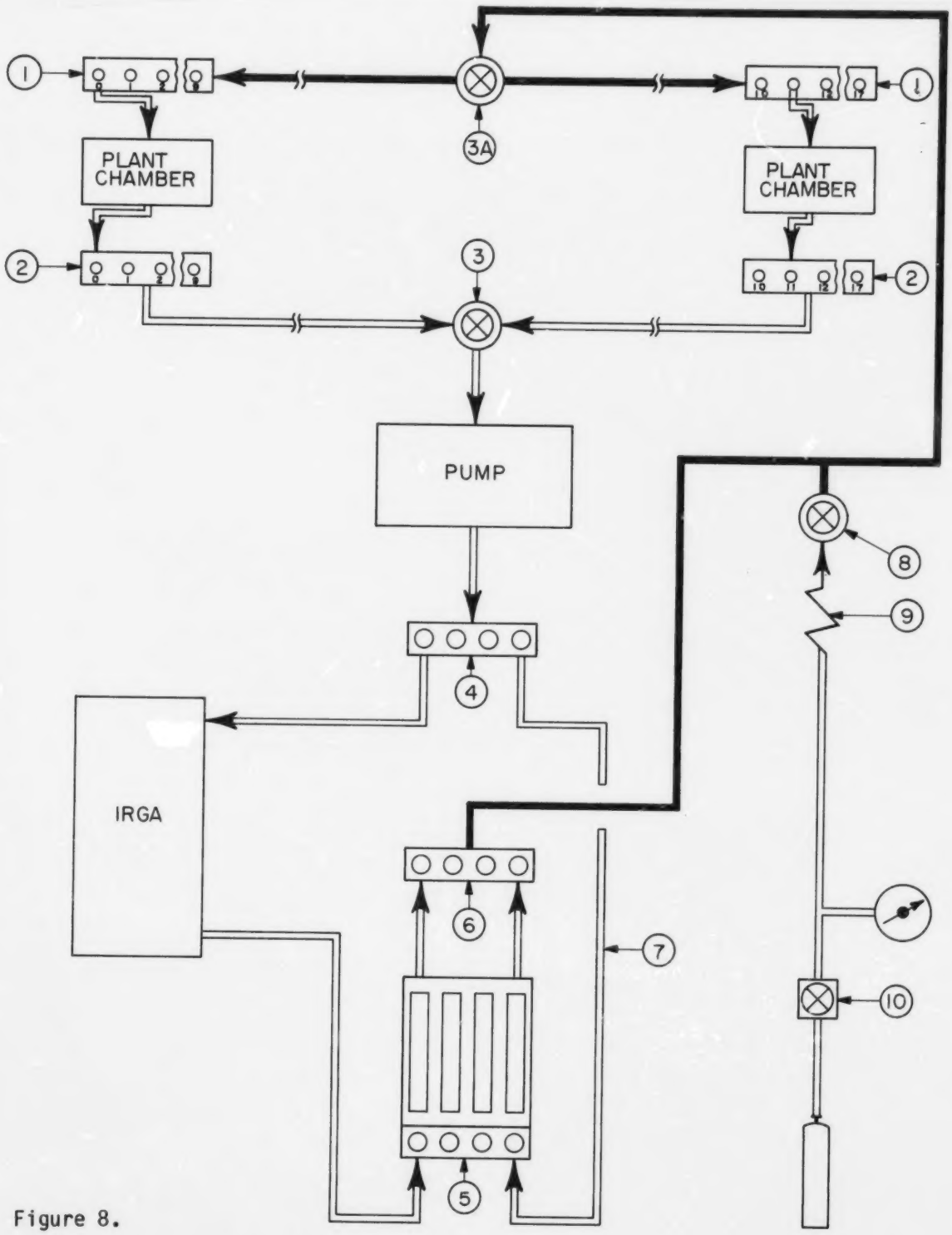


Figure 8.

The plant transpiration rate is determined by measuring the changing water pressure in a column (Fig. 9) which collects the transpired water condensing from chamber air. The slope is converted to ml of H_2O min^{-1} and represents the cumulative transpiration rate of all the plants in the chamber. The rate is calculated each time a measurement is made by determining the slope since the previous measurement and, if desired, is divided by the number of plants in the chamber to yield the average plant transpiration rate.

Under certain circumstances, there is a small amount of air movement in or out of the chamber. For instance, as the transpired water accumulates in the collector the chamber air volume decreases. This is reversed when the sampler is drained. Pressure equilibrium is maintained by a long, small-diameter tube with an activated charcoal filter at the end. These fluxes are small and do not significantly alter or influence the transpiration or photosynthetic rate measurements. Also, since they are filtered they are not a source for lab contamination.

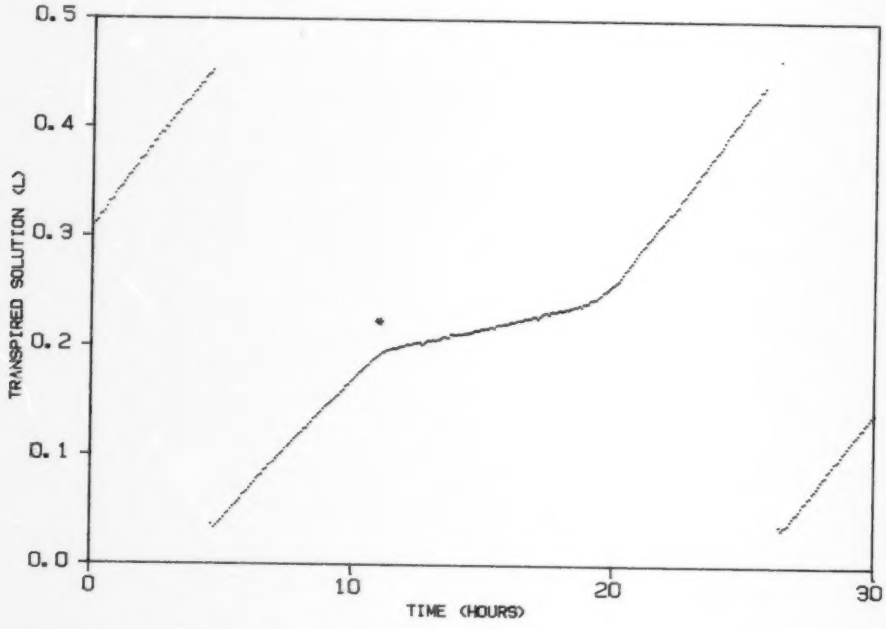


Figure 9. Transpiration rate measurement. This example of data starts by showing the slope of water collection (transpiration of plants) with the light on. At about 5 hours, the pot was full and was drained upon computer demand. At about 11 hours, the lights were turned off and the new slope represents the transpiration rate in the dark. The lights were turned on after 8 hours of darkness and the faster rate was quickly re-established. A computer program uses this information to calculate transpiration rate which is displayed in the summary statistics.

ROOT COMPARTMENT

HYDROPONIC SYSTEM. In this version of the plant exposure chamber, plants are grown in a hydroponic nutrient media that supplies all mineral nutrients and water needed for plant growth and development. Into this solution chemicals to be tested are introduced via a hypodermic injection port (Fig. 10) similar to that on a gas chromatograph. The same port is used to withdraw solution samples.

Chemicals contact various materials as the solution circulates through this system: plant containers and the solution reservoir both made of ultra high molecular weight, high density, linear polyethylene (Driscopipe); polyethylene fittings; polyethylene tubing (Beveline); and a Teflon/stainless steel pump. These materials were chosen because they are nonreactive with most chemicals and contain no plasticizers.

The solution is stirred and oxygenated to provide proper plant growth. Stirring replenishes the solution near the root surface with mineral nutrients, oxygen, and test chemicals. Since it is a closed system, oxygen is added in the form of compressed air via a capillary at the pump inlet. The flow rate of air is slightly in excess of the respirational O_2 demand of the roots (generally ~ 1 cm^3 min^{-1}). By using compressed air (20% O_2), the solution remains at air saturated equilibrium for the solution temperature (8.7 mg l^{-1} at 23C).

The solution level is monitored by a pressure transducer. The level is kept constant by measured additions which are made upon demand by the computer when a deficit is recorded. The volume is controlled to $\pm$ 15 ml (less than 0.3%).

Like the shoot container, the root container is not hermetically sealed. Small volume changes due to water loss by plant transpiration and CO_2 evolution from root respiration require gas exchange. This is accomplished by a small diameter tube with filters.

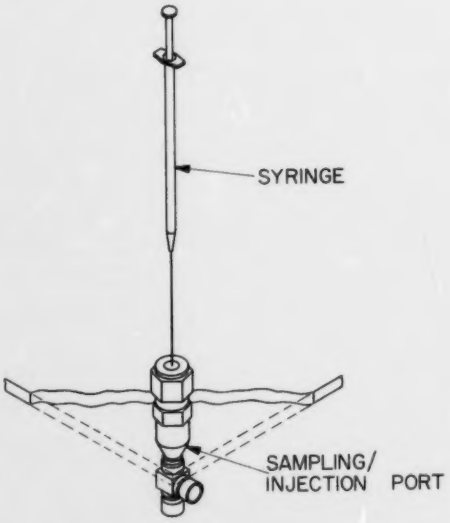
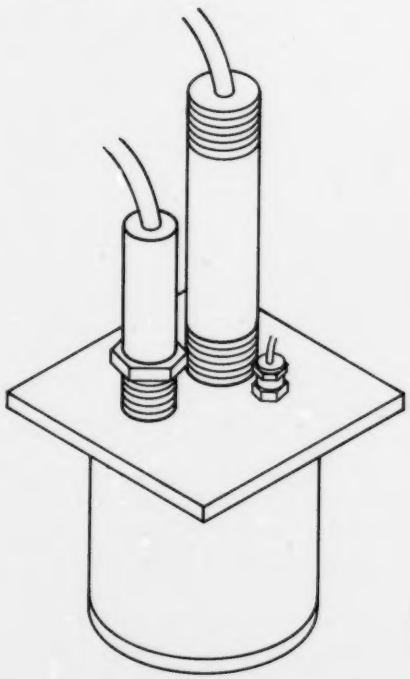


Figure 10. The solution sampling port is outside the CSTR and can be accessed without interrupting an experiment. The septum allows test chemicals to be added to the hydroponic solution and samples to be withdrawn using a hypodermic syringe.

Solution sampling for pH, temperature, and electrical conductivity are done electronically with probes mounted in a separate sampling pot (Fig. 11).

Several considerations have been included in the hydroponic system design to allow safe handling of toxic and/or radioactive chemicals. The solution quantity is as small as possible allowing just enough to fill the pots and cover the pump inlet. If the power goes off, the pots drain about 5 cm of solution into the mixing tank. Further draining is prevented by an anti-siphon safety valve. The capacity is large enough to contain all the solution in the plant pots if the anti-siphon fails. If the solution level control were to fail and the computer continuously demand more solution, a safety float switch disables the pump and valve at an intermediate level. When the power is off, the normally closed valve and unpowered pump preclude solution addition. A second normally closed valve at the pump precludes solution movement in case the valve at the chamber fails to seal. Also, there is a slight positive pressure (30-cm water head) towards the exposure chambers. If both valves and pump failed simultaneously, solution would flow only to fill the drain lines from the plant containers. No overflow could occur. The plumbing has been extensively tested for leaks under pressure. Additionally the plant carts are on an epoxy-sealed floor with a drain.

The test solution can be drained by a manual valve in the bottom of the mixing tank. The entire system can be rinsed with water, acid or other cleaning materials without injury to any component or possibility of spilling or leaking.



PROBE POT

Figure 11. The instrumentation pot is plumbed the same as a plant pot, but is located outside the CSTR. It contains a thermistor, electrical conductivity probe and a pH electrode.

Different plants utilize different amounts of mineral nutrients. Demands also change with time. This may result in an imbalance in the pH of the hydroponic media since the requirement for electrical neutrality is controlled by the abundance of H^+ and OH^- ions. In experiments where it is necessary to maintain a steady root environment the pH may be adjusted by the addition of HCl or KOH to the mixing tank. When the pH drifts from its programmed control level, the computer opens a valve to start a gravity feed of either acid or base until the control level is re-established. If needed, the volume of added amendments can be altered by changing the duration of injection.

SOIL SYSTEM. This version of the plant exposure chamber accepts potted plants which may be grown in soil or any other solid rooting media. The configuration of the chamber is similar to the hydroponic system, except that holes for soil pots (Fig. 12) replace the hydroponic system. The pots fit into holes in the table top and are sealed by a pressure-fitted closed cell foam. The soil is separated from the aerial compartment by plastic covers which fit over the pots and seal around the stem with closed cell foam. The soil compartment is provided with O_2 by drawing air through the cavity above the soil and filtering it before exhausting it outside. Irrigation is managed by the computer on the basis of time or determined by the amount of water lost by transpiration. Leachate is collected from each pot.

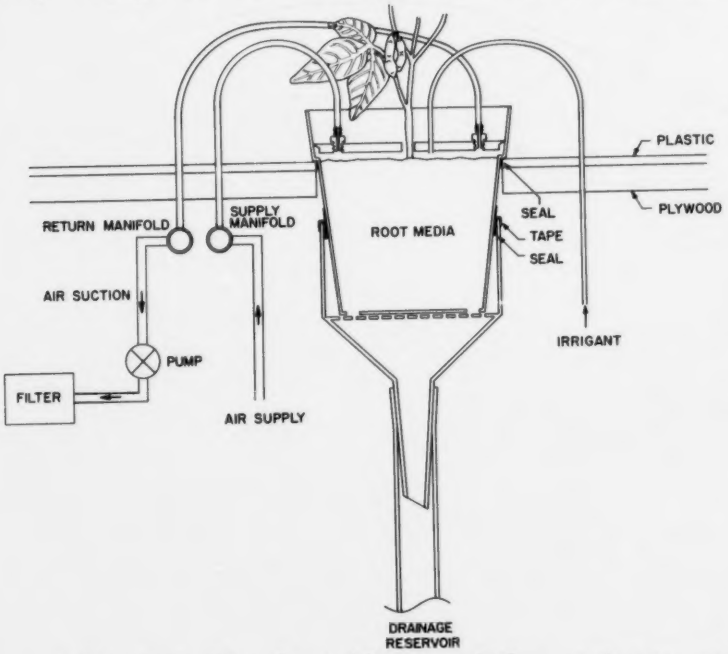


Figure 12. Soil pots fit in table top holes and are sealed in place by closed cell foam or putty. A drain system connects the pot to containers for collection of soil leachate. The soil in each pot is separated from the aerial compartment (CSTR) by a polyethylene plate which is sealed with teflon tape and polyethylene foam. The air space above the soil is continuously purged with air to provide oxygen and remove any volatile products. Exhaust gases are filtered and vented to the outside.

COMPUTER SYSTEM (CONTROL AND DATA COLLECTION)

HARDWARE. The electronic instruments and the automatic control functions of the Plant Exposure Laboratory are monitored and managed by a microcomputer (Apple //e). To the extent possible, the interfaces and other instruments are standard and readily available. However, because this is a new concept in laboratory automation, we designed and built most of the electronic hardware. A basic philosophy of the design process was to use as few different pieces as possible. Where practical, we chose to centralize circuits and share instruments. However, some instruments require close proximity to the sample and do not accommodate signal switching. Others require a constant power supply and are best adapted to local control. These criteria determined the instrument configuration. Table 1 lists the parameters measured and indicates whether the instrument was shared (location 1) or was housed on each chamber (2 & 3). Economic considerations were important in these decisions. For instance, the parts required to make a pH meter were inexpensive, easily assembled, and were thus attached to each chamber. Schematics and other details are included in Appendix 5. The computer and electronic interfaces and their functions are listed in Table 2.

Table 1. Measurement instrument locations and recorded units.

# ea	Parameter measured	Instrument manufacturer	Sensor	Location*	Signal to computer	Units recorded
5	Temp.	Bridge	Thermistor	1	+/-5VDC	°C
1	PAR[1]	Licor	Quantum	2	0-5VDC	$\mu mol\ s^{-1}\ m^{-2}$
2	Pressure	Bendix	Transducer	2	0-5VDC	ml H_2O
1	pH	Analytical Measurements	Electrode	2	+/-5VDC	pH units
1	EC[2]	Beckman	Electrode	1	0-5VDC	$dS\ m^{-1}$
1	CO_2 Flow Rate	Tylan	Gas flow Controller	3	0-5VDC	$mmol\ min^{-1}$
1	$[CO_2]$	Anarad	IRGA	1	0-5VDC	$mmol\ m^{-3}$
1	Humidity	Anarad	IRGA	1	0-5VDC	$mg\ m^{-3}$

*Location 1 is within the electronic equipment rack in the center of the lab area (see Figure 1).

Location 2 is in the electronic box for each exposure chamber.

Location 3 is mounted on the front panel of each exposure chamber and is plumbed with pressurized CO_2. It derives power and sends its signal through a plug-in connector on the electronic box.

1 Photosynthetically Active Radiation.

2 Electral Conductivity.

Table 2. Computer specifications and electronic components.

Item	Manufacturer	Specifications and Function
Computer	Apple //e	128 K memory
A/D converter	IS* AI13	12-bit analog input system. 16 separate input channels with various full-scale ranges.
I/O	IS* DI09	32-line digital I/O interface.
Switching	Constructed	Electronic multiplexer switching unit connected to the DI09. Selects desired chambers for electronic measurements or control and selects gas sampling manifold for desired chamber.
	IS* UI16	16 completely isolated AC or DC relays connects to the DI09.
Monitor	Zenith	Monitor
Modem	Hayes	Micro modem, 300 baud. Used to alert investigators of mechanical problems or power failure and allows remote interrogation of conditions and data.
Disc	Corvas	44 Mega byte. Used to store programs and data.
	Apple	5-1/4 inch floppy disc. Used to enter program changes and to remove data.
Thermistor interface	Constructed	Provides signal, amplifier and resistance network for measuring temperature with thermistors.
Chamber Electronics	Constructed	Provides for measurement of pH, light, hydroponic solution level, transpired water level. Electrical conductivity, signal switching, adjustable voltage for fan and impellar speed control, solenoid drivers, digital/analog converter for CO_2 control.

*IS = Interactive Structures, Inc.

SOFTWARE. The programs were written in Pascal (implemented in Apple Pascal 1.2) and include the major routines listed in Table 3. A detailed description of the programs is found in Appendix 1. The Pascal code is contained in Appendix 2.

Two sampling routines operate simultaneously. The first is a continuous cycle which samples chamber gases for CO_2 and H_2O. It is sequenced to the next function after a pre-defined interval. The interval (30 seconds) is sufficient to fill the measurement system with chamber air and yield a stable response. The length of the sampling cycle depends on the number of chambers being measured.

The second routine is activated periodically (generally every 5 minutes) and interrogates the sensors at each chamber. The computer selects the first plant exposure chamber by sending a signal which latches a series of relays. In this condition, the chamber sensors are connected to the computer's analog to digital (A/D) converter. A 5-second delay to equilibrate the sensors follows, and then the outputs are read, calibration corrections made and the data are stored in the appropriate engineering units in an array which identifies the chamber and the sampling time. The computer then disconnects and repeats the process until each chamber in the lab has been interrogated.

Table 3. Program routines and functions.

Routine	Function
Control	Operates the sampling interupts, collects data, manages the controlled features.
Examine	Provides a graphic display of data. The output can specify any parameters and over any time period.
Statistics	Determines the mean, standard deviation, high/low points, and slope for each measured parameter during a defined interval.
Modem	Evaluates environmental parameters against defined limits and calls a list of telephone numbers until someone acknowleges the emergency condition. This program also facilitates interrogation of data files and allows control of certain conditions from remote sites.
Parameters	Provides entrance of chamber and experimental variables which are used by the other routines to evaluate, calculate and determine subsequent action.

Data which fall outside pre-defined limits are flagged. The computer uses those flags to return to the appropriate chamber and correct the aberrant condition. Again, the chamber is selected by latching the connect relays. The conditions listed in Table 4 then can be managed. Finally the computer waits until the next sampling time.

Table 4. Automatically controlled functions on each plant exposure chamber.

Function	When and How
Hydroponic	When a deficit in the solution level is detected, two valves are opened, one on the selected chamber and another at the hydroponic reservoir. A solenoid pump is pulsed a pre-defined number of strokes to deliver a measured amount of solution to the chamber in need.
Irrigation	Based on time or a defined amount of transpiration, the system for delivering hydroponic solution can be used to deliver a measured amount of solution to potted plants.
pH	When the pH of the hydroponic solution is outside the defined limits, a valve is opened to allow addition of either acid or base. This is accomplished by gravity feed through a small tube of either 0.1 N HNO_3 or KOH for a defined time (generally 2-3 seconds) calculated to adjust the pH to within acceptable limits.
CO_2 injection rate (Photosynthesis)	CO_2 continuously flows into the chamber during the light period to balance the CO_2 assimilated by the plants. When the CO_2 concentration is outside the defined limits, an adjustment is made by sending a digital signal to an A/D converter at the selected chamber, which generates the control voltage to the gas flow controller. The logic and program steps of this adjustment are found in Appendix 2.
Transpired water collector	When the pressure sensor indicates the collection tube is full, a valve is opened to drain the transpired water into a sump or sample bag. The valve is closed on the next measurement cycle and collection continues.
Automatic sampling	Based on time or amount of transpired water, an automatic sampler is activated (electronic components are connected but sampler is not yet in use). The sampler will collect a measured amount of transpired water and either the hydroponic solution or the soil leachate. A temporary lid (stainless steel ball) will be placed when the fraction collector is advanced.

Data are partially analyzed during collection. A statistical routine determines the mean, standard deviation, high/low points, and slope for each measured parameter during a defined interval, typically the light or dark period of each day. This summary information is always available; it also is stored with the detailed record. Currently we evaluate the detailed record only if the summary statistics indicate a problem or if the experiment requires that information.

HYDROPONIC NURSERY

Mature plants are needed for experiments on the uptake, translocation, accumulation and biodegradation of toxic organic chemicals. The plant exposure chambers are specifically designed to conduct such studies, but plant growth takes too long for that purpose. Therefore a plant nursery (Fig. 13) was built to support uptake studies. Plants need to be healthy, have a root geometry compatible with the exposure chambers, and be individually supported.

The hydroponic plant nursery is set up in a glasshouse which is programmed to start cooling above 21C and heating below 15C. In winter incident radiation is supplemented with 400-watt high pressure sodium vapor lamps for 16 hours. The nursery supplies water, mineral nutrients, and oxygen to the root. The solution flow rate is equivalent to one solution change every 1.3 minutes. Oxygen is dissolved in the solution by contact with the air during its return to the nutrient reservoir.

The nutrient solution, a modified Hoaglands, is supplied to the mixing tanks under pressure and the level is controlled by a float valve. A parts list and construction details are included in Appendix 4.

PERFORMANCE

CSTR. The container which covers the aerial parts of the plants was designed with the characteristics of a continuously stirred tank reactor (CSTR). Most reactor tanks are not closed and include an inlet and outlet where reaction rates are determined by measuring the difference in concentration or chemical state between those points. In these closed tanks rates, are measured by determining the rate of extraction (transpired water) or injection (CO_2) required to maintain a steady state. A condition implicit in the CSTR concept is that the gas is uniform throughout the chamber. Although this "ideal mixing" condition is impossible to achieve, it is approachable. Another condition important in plant studies is the requirement for sufficient air turbulence at the leaf surface to prevent stagnant conditions so that plants respond to the boundary layer rather than the bulk air mass.

MIXING. The mixing conditions were tested by determining the time required for a chamber to reach a stable CO_2 concentration after an instantaneous injection of CO_2. The air sampling system consists of tubes, valves, a pump, and the volume of the IRGA, all of which must be considered in determining the chamber mixing rate. The data in Figure 14 were used to determine the mixing rate.

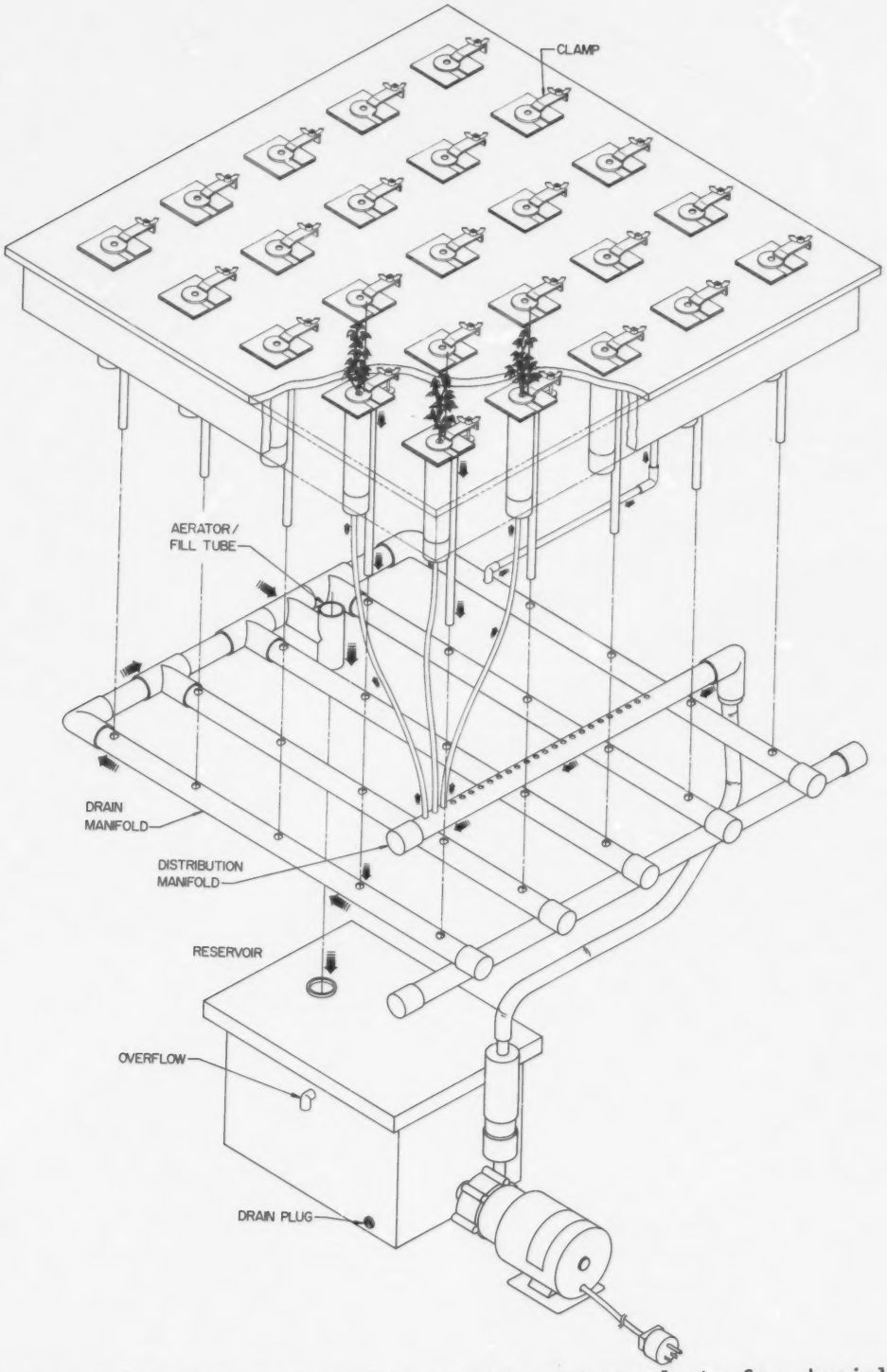


Figure 13. Hydroponic plant nursery provides mature plants for physiological studies in the exposure chambers. The pot dimensions are similar and the table is free of any obstructions so that transplanting is easy.

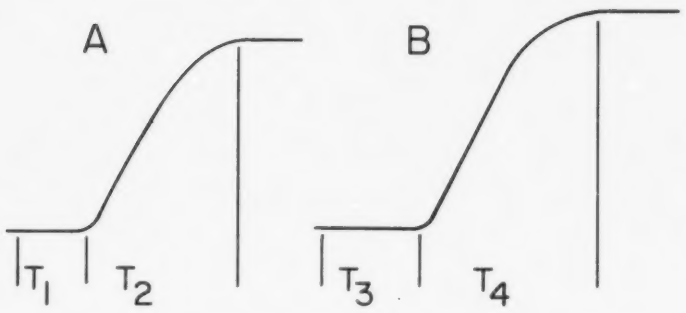


Figure 14. CSTR mixing rate. (A) The CO_2 concentration changed in response to instantaneously changing the air source at the chamber sampling inlet. The time (T_1 = 4.2 sec) represents the period required to purge the sample plumbing, manifolds and pump. The time (T_2 = 9.0 sec) represents the IRGA purging time and delay in the electronic response.

(B) The CO_2 concentration changed in response to injecting 50 cm^3 of CO_2 into a stable (empty) plant exposure chamber. The time ($T_3 - T_1$ = 1.5 sec) represents the period from injection until some of the injected gas reached the sample inlet. The time $(T_3 + T_4) - (T_1 + T_2)$ represents the chamber mixing time.

At a median impeller speed (500 rpm), the time to completely mix an introduced gas in the chamber was about 2 seconds. This mixing rate is fast compared to any changes expected from treatments, thus ideal mixing is assumed in determining all gaseous rate constants.

TURBULENCE. Air turbulence is created by an impeller mounted in the top of each chamber. Impeller speed is adjusted by setting the voltage to the motor. Turbulence was measured at three heights in an empty chamber as a function of impeller speed. In a planted chamber (Fig. 15) the response was similar, somewhat moderated by the plant canopy.

PHOTOSYNTHESIS. Control of CO_2 concentration in a closed chamber requires that the rate of injection equal the rate of plant CO_2 assimilation. This is accomplished by a computer routine which operates valves and a gas flow regulator and works in the following manner: the CO_2 concentration in each chamber is measured sequentially by an IRGA. At the time of each measurement, the value is compared to the desired control value. From memory the computer gets the previously measured value, the time since last measurement, and the current rate of CO_2 injection.

It applies a logic routine (Appendix 2) to make the necessary correction and records the rate of photosynthesis.

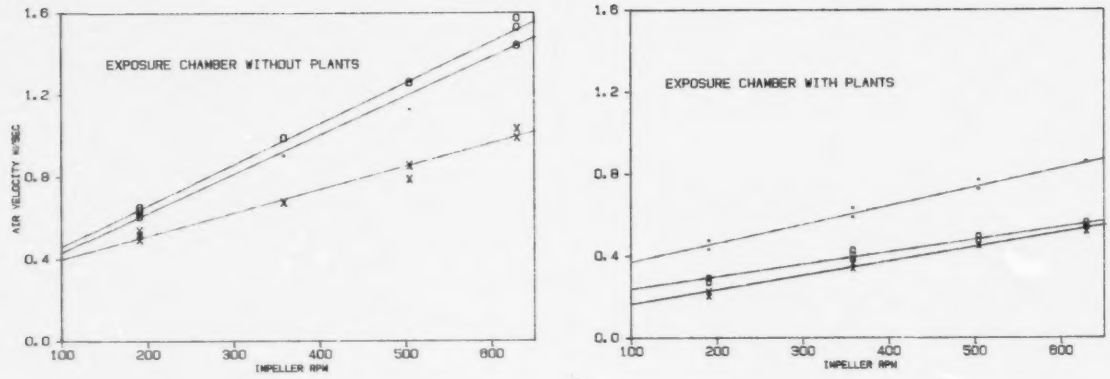


Figure 15. Air turbulence in the CSTR is a function of impeller speed. Air velocity was measured with an omni-directional hot wire annemometer at 15 cm (X), 30 cm (0), and 60 cm (.) above the table top at various impeller speeds.

A simple response test (Fig. 16) of this control system included two chambers, one with barley, the other with soybeans. The selected concentrations were changed after a plateau was reached. The test sequence included stepping from high to low, then low to high, a static control, and finally with the control system off, a compensation curve was recorded.

ENVIRONMENTAL AND PLANT CONDITIONS

The computer records of plant functions and environment may be displayed graphically (Fig. 17) for any of the data sets collected. Summary tables are also available for each day or night period (Table 5). Both presentations are available at all times in the control lab or by modem, but since the graphs take much more time, the summary tables are the first indication of satisfactory plant functioning and acceptable chamber performance.

Figure 16 (page 23). Automatic CO_2 control is shown for two plant exposure chambers filled with barley (odd numbers) and soybeans (even numbers). Different CO_2 concentrations were achieved by entering the desired value into the computer. The first 8-hour test demonstrates the control which will normally occur. The next series (3 & 4) demonstrates adaptation time with increasing concentrations and (5 & 6) with decreasing concentrations. The final series shows the CO_2 compensation curve created with the control system turned off and no CO_2 is provided to the chamber.

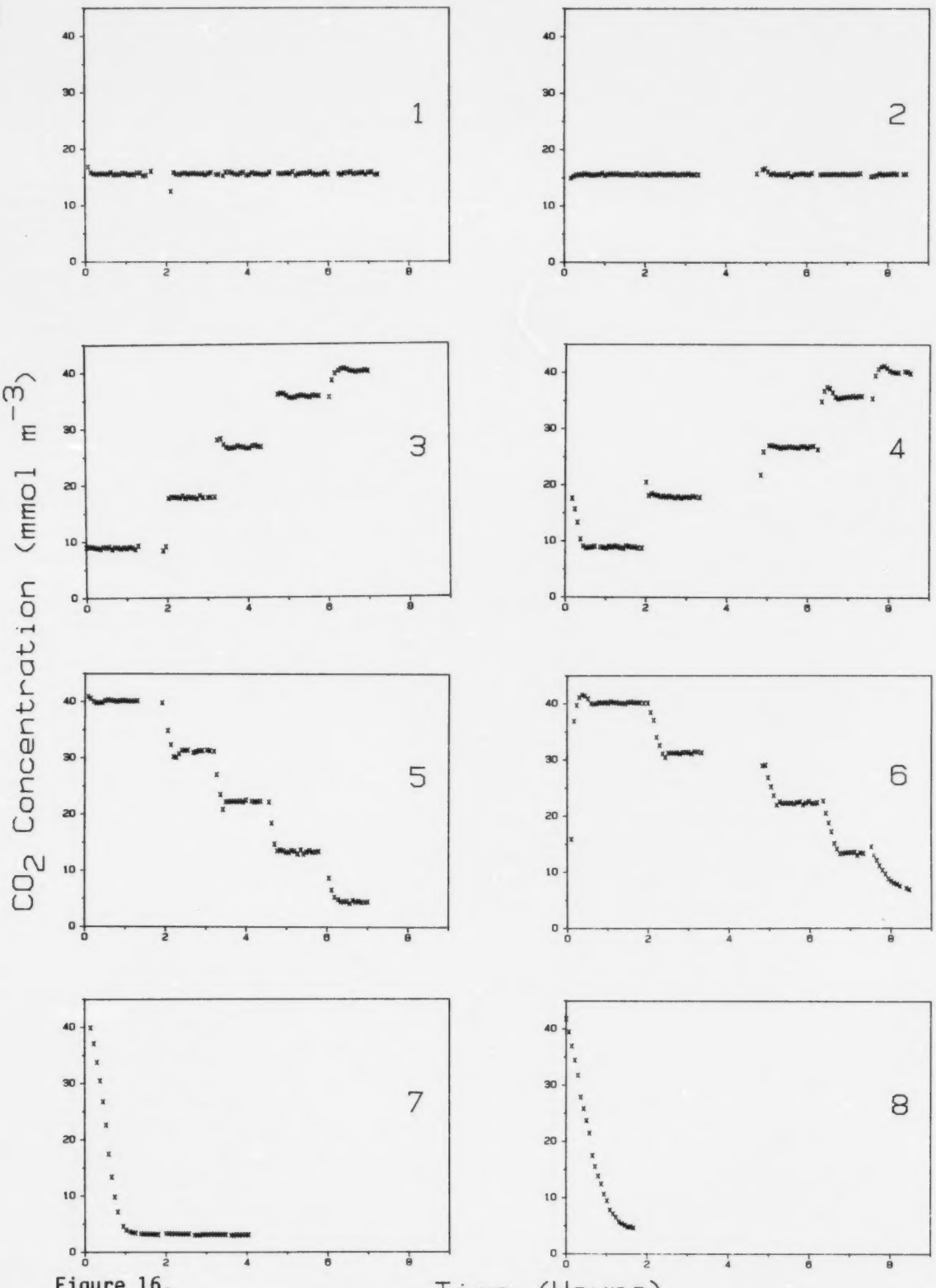


Figure 16.

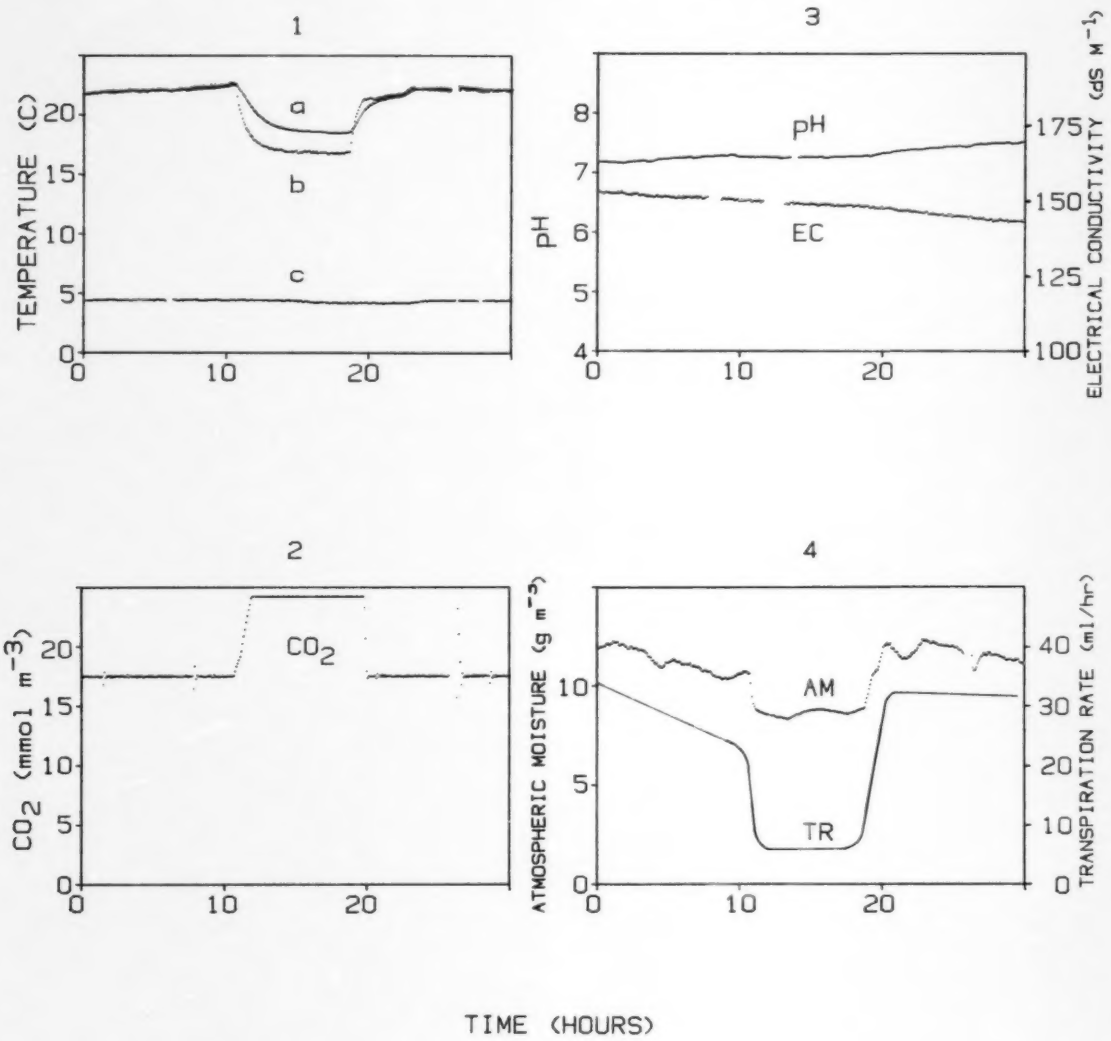


Figure 17. Computer display of measured parameters in one chamber over 30 hours. [1] Temperatures for (a) Hydroponic solution, (b) chamber air, and (c) condenser. [2] CO_2 concentration in chamber air. Control level was set at 17.8 mmol m-3 (400 ppm) and uncontrolled during the dark. [3] Nutrient solution pH and electrical conductivity (EC) were uncontrolled in this example. [4] Atmospheric moisture (AM) and transpiration rate (TR).

Table 5. Statistics screen display. This table presents data from a plant exposure chamber which is collected on channels 1 - 16 of the A/D converter and channels 17, 18 which are calculated values. All values are expressed in engineering units which correspond to the measurements made by that sensor. Because of space, units for each value are not identified on the display. "N" indicates the number of measurements which constitute the mean (Mean), the standard deviation (SD), and coefficient of variation (CV) in a set of measurements. An alternate presentation is used when a rate is calculated (i.e., transpired water). In that case the slope (a), intercept (b), correlation coefficient (r), and standard error (SE) are displayed in the same spaces. Maximum (Max) and minimum (Min) values are displayed to allow the operator to evaluate any abberant conditions. The period of data accumulation, indicated in the upper right, is limited to day or night, periods which will generally be expected to be constant, and transition, when the system is changing from light to dark or vice versa. Other header information is common to all main menu displays and is explained in the text.

Time: [9:39:36] Date: [15 Jan 85] Memory: [7090] Record: [6315]
Air system [purging] Stats [+] Data [+] Regular [+] Air [+] Phone [-]
Position [7] Cart [11] Date [1.1585] Period [day]

Chan	N	Mean/a	SD/b	CV/r	Max/SE	Min
[0]	[53]	[4.35]	[0.08]	[1.80]	[4.45]	[4.20]
[1]	[53]	[22.90]	[1.43]	[6.22]	[24.06]	[20.18]
[2]	[53]	[23.37]	[0.83]	[3.54]	[24.25]	[21.86]
[3]	[53]	[22.05]	[0.66]	[3.00]	[22.57]	[20.65]
[4]	[0]	[ ]	[ ]	[ ]	[ ]	[ ]
[5]	[53]	[5.16]	[0.05]	[0.95]	[5.26]	[5.12]
[6]	[53]	[1.74]	[0.04]	[2.41]	[1.78]	[1.65]
[7]	[53]	[6.54]	[0.03]	[0.51]	[6.62]	[6.47]
[8]	[53]	[0.03]	[0.01]	[34.16]	[0.05]	[0.02]
[9]	[53]	[0.00]	[0.00]	[14.44]	[0.00]	[0.00]
[10]	[53]	[81.60]	[3.60]	[4.41]	[86.04]	[76.53]
[11]	[53]	[386.95]	[58.50]	[15.12]	[484.44]	[345.11]
[12]	[53]	[16.16]	[2.72]	[16.83]	[18.11]	[6.86]
[13]	[0]	[ ]	[ ]	[ ]	[ ]	[ ]
[14]	[0]	[ ]	[ ]	[ ]	[ ]	[ ]
[15]	[0]	[ ]	[ ]	[ ]	[ ]	[ ]
[16]	[53]	[161.89]	[72.86]	[45.00]	[300.00]	[90.00]
[17]	[53]	[0.00]	[0.00]	[0.00]	[0.00]	[0.00]
[18]	[0]	[ ]	[ ]	[ ]	[ ]	[ ]

<--/--> Select Screen; R)eturn to Pascal; Error [0]

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